Docket No.: 20555/1203301-US3
(PATENT)

IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re Patent Application of:
Daniel G. Chain

Application No.: 10/084,380 | Confirmation No.: 3496

Filed: February 28, 2002 | Art Unit: 1649

For: **SPECIFIC ANTIBODIES TO AMYLOID BETA PEPTIDE, PHARMACEUTICAL COMPOSITIONS AND METHODS OF USE THEREOF** | Examiner: K. A. Ballard

DECLARATION OF HOWARD J. FEDEROFF, M.D., PH.D., UNDER 37 C.F.R. §1.132

MS RCE
Commissioner for Patents
P.O. Box 1450
Alexandria, VA 22313-1450

Dear Sir:

Howard J. Federoff declares and states as follows:

1. I am a citizen of the United States, more than twenty-one years of age, and make this Declaration in support of this application.

2. On April 1, 2007, I assumed the position of Executive Vice President for Health Sciences and Executive Dean of the Georgetown University Medical Center. Prior to joining Georgetown I served as the Senior Associate Dean for Basic Research and Professor of Neurology, Medicine, Microbiology and Immunology and Professor of Oncology and Genetics at the University of Rochester School of Medicine. I joined the Rochester faculty in 1995 as Founding Director of the Division of Molecular Medicine and Gene Therapy in the Department of Neurology. In 1997, I was appointed founding Director of the Center for Aging and Development Biology at the Institute of Biomedical Sciences. I was appointed Director of the University of Rochester Neuroscience Program in 1997. In 2006 I was named Chairman of the NIH Recombinant DNA Advisory

Committee (RAC). I am a physician Board Certified in Internal Medicine and Endocrinology and Metabolism.

3. I have conducted research in the field of gene therapy for neurodegenerative diseases, including Alzheimer's disease, since 1988. Specifically, I have studied the mechanisms of human neurodegenerative diseases and have developed gene therapeutic strategies that are progressing towards clinical trials. I founded an NIH supported Parkinson's Disease Gene Therapy Study Group, which I directed from its inception in 2000 through December 2006 and to which I remain a member. This Group is anticipating a pre-Investigational New Drug Application meeting with the U.S. Food and Drug Administration in the fall of 2007 in connection with a Parkinson's AAV-based gene therapy product, further details of which are confidential.

4. I am familiar with the etiology, diagnosis and treatment of Alzheimer's Disease through my medical training, my research on the early neuroinflammatory phase of Alzheimer's Disease, the development of an active vaccine for Alzheimer's Disease and studies on antecedent and early peripheral biomarkers for Alzheimer's Disease. My work on antecedent biomarkers for the diagnosis of Alzheimer's disease has lead to an ongoing clinical study. In this regard, I designed a prospective molecular epidemiological study in which individuals at high risk for Alzheimer's Disease are recruited into our cohort, whereupon they undergo detailed neuropsychological examination and blood studies. When subjects present mild cognitive impairment or frank Alzheimer's Disease their leukocyte biomarkers are analyzed for transcription and protein expression patterns that are signatures indicative of pre-symptomatic Alzheimer's disease.

5. I am a co-inventor of four U.S. patents related to heterologous expression of mammalian genes in transgenic animals, including U.S. Patent No. 6,051,428, entitled "Rapid Production of Autologous Tumor Vaccines." This patent discloses using herpes simplex virus amplicons containing genes encoding for immunomodulating proteins to transduce tumor cells with high efficiency either *ex vivo* or *in vivo*.

6. A copy of my curriculum vitae is attached as Exhibit A.

7. I am not a co-inventor of the subject patent application and have no financial or business interest in Intellect Neurosciences, Inc., a company that I understand has rights in this patent application.

8. I have been asked for my opinion on whether the specification of provisional application 60/041,850, filed April 9, 1997 ("the '850 application"), would have enabled one of ordinary skill in the art to use gene therapy to inhibit neurotoxicity of β-amyloid protein ("Aβ") by contacting soluble Aβ in the cerebrospinal fluid (CSF) of a patient suffering from Alzheimer's Disease with a free-end specific antibody to Aβ at the time the application was filed. This opinion is based on the information disclosed in the '850 application, as it would have been understood by one of ordinary skill in the field of the application as of April 9, 1997, the filing date of the '850 application and the techniques that were in use in the field of gene therapy as of the filing date of the '850 application.

9. In forming my opinion, I considered the '850 application, the Office Action for the subject application mailed September 13, 2006, the pending claims that were filed on June 30, 2006, and the state of the art in the field of gene therapy as of the April 9, 1997 filing date of the '850 application, particularly as applied to gene therapy for disorders of the nervous system.

10. Based on my review of the '850 application and my education and experience in the field of gene therapy, I conclude that no later than April 9, 1997, the information disclosed in the '850 application and the techniques that were then well known to those working in the field of gene therapy would have been sufficient to enable a person of ordinary skill in the field of gene therapy to use gene therapy to practice the methods called for in the claims of the subject patent application, for inhibiting accumulation or neurotoxicity of Aβ by contacting soluble Aβ in the CSF of a patient suffering from Alzheimer's Disease with a free-end specific antibody to Aβ.

11. My first reason for arriving at this conclusion is that by April 9, 1997 (the filing date of the '850 application) the level of skill in the art of gene therapy for the nervous system was extremely high. The systematic re-engineering of viruses to be deployed as vectors for gene transfer evolved greatly in the early to mid-1990s with the derivation of a number of virus vectors,

notably Adeno-associated virus (AAV) vector (*see*, Nahreini et al., 1993, *Gene* 124:257-262, attached as Exhibit B) which was suitable for human gene therapy (*see* Kaplitt et al., 1994, *Nature Genet.* 8:148-154, attached as Exhibit C). By April 1997, gene therapy by delivery of genes to the nervous system had been used to treat glioblastoma multiforme in mammals (Eck et al., 1996, *Hum. Gene Ther.* 7:1465-1482, attached as Exhibit D). Thus, by the filing date of the '850 application the level of ordinary skill in the art of gene therapy was at an advanced stage.

12. My second reason for arriving at the conclusion that the '850 application would have been enabling for gene therapy at the time the application was filed is that the level of understanding of Alzheimer's disease, the level of skill in the art of gene therapy for neurological diseases, and the amount of guidance in the '850 application provide a rational, predictable basis for using the methods for treating Alzheimer's called for in the claims pending in this application. In particular, a successful gene therapy protocol can be predicted by satisfying the following four criteria:

i. An understanding of the disease mechanism;

ii. Selection of a therapeutic gene with action on a target component that is necessary for the pathogenesis of the disease;

iii. Selection of an appropriate virus vector that can deliver the therapeutic gene to the required anatomical area and express levels of the therapeutic gene sufficient to act on the target for the duration of disease; and

iv. A means to deliver the therapeutic gene to the patient.

13. By the filing date of the '850 application each of the criteria set forth in paragraph 13 had been fulfilled, as follows:

i. Mechanism of Alzheimer's Disease. It had long been postulated that Alzheimer's disease was caused by Aβ peptides that are derived from the amyloid precursor protein (APP). By April 1997, the cognitive impairment observed in Alzheimer's disease was thought to derive, at least in part, from Aβ assemblies interfering with synaptic neurotransmission.

Aβ peptides also form insoluble neuritic plaques that accumulate in the brains of Alzheimer's patients. Although the precise mechanism by which Aβ peptides contribute to the pathology of Alzheimer's disease continues to be investigated, by the filing date of the '850 application, a causal relationship between Aβ peptides and Alzheimer's disease had been established, by the discovery that a major form of familial Alzheimer's disease was caused by mutations in the gene encoding the APP (the "Swedish mutation") that leads to increased formation of Aβ peptides. *See* Citron et al., 1992, *Nature* 360:672-674, attached as Exhibit E. The finding that transgenic mice bearing the "Swedish mutation" developed Alzheimer's-like pathophysiology, including cognitive impairment and neuritic plaques, confirmed a pathogenic role of the Aβ peptides. Hsiao et al., 1996, *Science,* 274:99-103, attached as Exhibit F.

ii. Selection of therapeutic gene with action on a pathogenic component. By the filing date of the '850 application, those of ordinary skill in the art identified reducing the production of pathogenic Aβ peptide and accelerating the clearance of the processed and parenchymal Aβ peptides as means to treat Alzheimer's disease. *See review by* Cordell, B., 1994, *Ann. Rev. Pharmacol. Toxicol.* 34:69-89, attached as Exhibit G. By 1996 in vitro studies had demonstrated that monoclonal antibodies inhibit fibrillar aggregation of Aβ peptides. Solomon et al., 1996, *Proc Natl. Acad. Sci. USA* 93:452-455, attached as Exhibit H. These results established to those skilled in the art that a gene therapeutic, particularly an antibody, that binds to Aβ peptides and promotes their metabolism and clearance was a viable Alzheimer's disease therapy.

iii. Virus vector for delivery of Alzheimer's disease therapeutic gene. By the filing date of the '850 application, vector platforms for gene therapy in the nervous system had been developed, including those based on adenovirus, herpes simplex virus, retrovirus, lentivirus and AAV, a parvovirus. By the filing date of the '850 application, those skilled in the art appreciated that AAV provided certain advantages for neurological gene therapy. Thus, it was well known to those working in the field of gene therapy that AAV does not integrate into neurons, infects central nervous system tissue (i.e., is neurotropic) when

packaged into serotype 2 capsids, does not promote a sustained immunological response that limits gene expression, and achieves the long term gene expression that is required for treatment of progressive neurological diseases, such as Alzheimer's disease. Kaplitt et al., *supra*, Exhibit C. Thus, by the filing date of the '850 application, one skilled in the art would have identified AAV as a preferred vector for neurological gene therapy.

iv. Means for delivery. By the filing date of the '850 application, neurosurgical approaches to several intracranial maladies had required the use of stereotaxy to precisely target and deliver therapeutic agents to the brain. Thus, stereotactic surgery using conventional frames, fiducials (i.e., markers for precisely locating anatomical structures in three-dimensional space using brain images obtained by CT or MRI), and imaging was a standard procedure being practiced at many neurosurgical programs throughout the world. The intracranial delivery of a vector-containing fluid to an anatomically circumscribed region of the brain was familiar to practitioners with training in functional neurosurgery. *See, e.g.*, Bouvier et al., *Appl. Neurophysiol.* 1987, 50:223-226 (attached as Exhibit I) and Siegfried, J., 1993, *Acta Neurochir. (Wien)* 124:14-18 (attached as Exhibit J). Thus, by the filing date of the '850 application, one of ordinary skill in the art could have delivered a vector encoding a therapeutic antibody to the brain of an Alzheimer's patient.

14. My third reason for concluding that the '850 application is enabling for gene therapy to treat Alzheimer's disease is that the '850 application provides an abundance of guidance for production of an end-specific antibody to Aβ for use in a treatment for Alzheimer's disease. By following the guidance given in the '850 application and using techniques commonly practiced by gene therapists and molecular biologists in April 1997, I (or one of my post-doctoral students with several years of laboratory research experience in the field of gene therapy) could have used the teachings of the '850 application to practice the claimed methods for treating Alzheimer's disease, as follows.

15. First, the '850 application provides extensive guidance for generating end-specific antibodies to Aβ (*see* '850 specification at pages 30-32) and testing the specificity of the end-specific antibodies and their in vitro efficacy in blocking Aβ aggregation and Aβ-induced

cytotoxicity (*see* '850 specification at pages 33-36). The '850 application further provides detailed guidance for isolating the immunoglobulin genes that encode end-specific antibodies, including a method for preparing single chain fragments (scFvs) (pages 37-38). As set forth in the '850 application, the end-specific scFvs would encode a small, secreted antibody that can diffuse within the interstitial space of brain and CSF, where it would contact and bind to Aβ peptides ('850 application at pages 16-21). The formation of scFv-Aβ complexes would reduce the Aβ peptides pathogenic potential and promote clearance of Aβ from the CSF. These methods to make scFvs were standard in the field by early 1990s and could have been successfully practiced by those with ordinary skills in molecular immunological methods, which I consider to have been among the skills well known to one of ordinary skill in the field of gene therapy in April 1997. *See, e.g.,* Colcher et al., 1990, *J. Natl. Cancer Inst.* 82:1191-1197, attached as Exhibit K.

16. The '850 application further provides detailed guidance for choosing a vector and producing constructs that will reproducibly express a scFv transgene in neuronal tissue to provide end-specific anti-Aβ antibody to the CSF. Thus, the '850 application identifies AAV as a vector of choice for expressing free end-specific antibodies in neuronal tissue (page 24, line 20 through page 25, line 25). AAV vectors were widely used by the filing date of the '850 application. *See, e.g.,* Nahreini, *supra,* Exhibit B; Kaplitt, *supra*, Exhibit C; Goodman, et al., 1994, *Blood* 84: 1492-1500, attached as Exhibit L; and McCown, et al., 1996, *Brain Res.* 713: 99-107, attached as Exhibit M. The '850 application further discloses that a leader or signal sequence should be appended to the recombinant antibody gene, to allow secretion of antibody from expressing cells (page 21, lines 24-27). At the time the '850 application was filed, it would have been routine to append a mammalian signal sequence to the N-terminus of a recombinant antibody. The '850 application further identifies the Thy1 promoter as a preferred promoter for obtaining expression of scFvs in the brain (page 23, lines 8-12) and identifies the pSSV9 vector as a vehicle for assembling the Thy1-scFv construct, including assembly of the Thy1-scFv with the additional sequences required for efficient expression, e.g., viral inverted terminal repeats (ITRs) and SV40 early region polyadenylation signal (page 39, lines 1-20). The methods used to prepare the Thy1-scFv constructs and clone them into AAV were standard methods, used routinely in molecular biology laboratories. *See, e.g.,* Nahreini et al., *supra*, Tab B; Flotte et al., 1993, *Proc. Natl. Acad. Sci. USA*

90:10613-10617, attached as Exhibit N; and Walsh et al., 1992, *Proc. Natl. Acad. Sci. USA* 89:7257-7261, attached as Exhibit O.

17. The '850 application further provides guidance for obtaining a high titer recombinant virus stock to be administered to an Alzheimer's patient. Thus, the application describes a method for packing the Thy1-scFv-containing vector and culturing virus to prepare a viral stock (pages 39-41). These procedures were routine at the time the '850 application was filed and would yield recombinant virus of sufficient concentration and purity suitable for stereotactic injection into the brain of human subjects. The methods described in this section were practiced in a number of viral vector and virology laboratories throughout the world by 1997. *See*, e.g., Nahreini, *supra.*

18. The '850 application further describes methods for delivering the recombinant Thy1-scFv to cells of the central nervous system (pages 23-27 and 44). These methods include the direct intracerebral stereotactic delivery of the therapeutic rAAV type 2 virus vector into the brain (page 27, lines 7-13 and page 44, lines 7-10). The methods described in the '850 application relating to stereotactic injection were in common practice by 1997. *See, e.g.,* Bouvier G, et al., *supra.*; Lindvall et al., *supra.*; Siegfried, *supra.* Injection of recombinant virus would produce localized infection of neuronal tissue (Kaplitt et al., *supra.*), followed by expression of the scFv transgene, and secretion of the end-specific antibody into the CSF, where it would diffuse and bind Aβ peptide. In particular, Aβ end-specific antibodies would distribute throughout the interstitial space and CSF, where the majority of Aβ peptides reside in patients with Alzheimer's disease.

19. In short, as outlined in paragraphs 16-18, the '850 application provides guidance that would allow one of ordinary skill in the art in April 1997 to:

a. generate free end-specific antibodies to Aβ;

b. prepare recombinant single chain versions of such antibodies;

c. place the recombinant antibody genes under the control of an appropriate promoter and other sequences required for expression and secretion of

recombinant antibody in neuronal cells and in an appropriate viral vector;

d. prepare a high titer viral stock; and

e. introduce recombinant virus into the nervous system, where it would infect neuronal cells, which would then express and secrete the end-specific antibody into the CSF.

20. As set forth in paragraph 14 above, the recombinant end-specific antibody would then bind Aβ and lead to a reduction in assembly of Aβ or clearance of Aβ from the CSF. Thus, in my opinion, in April 1997 the '850 application would have provided sufficient information to allow one of ordinary skill in the art to practice the claimed methods of inhibiting accumulation or neurotoxicity of Aβ by contacting soluble Aβ in the CSF of a patient suffering from Alzheimer's Disease with a free-end specific antibody to Aβ without undue experimentation. No further information beyond that given in the specification would have been needed to enable a worker in the gene therapy field to practice the methods called for in the presently pending claims.

21. I do not agree with the Examiner's conclusion that gene therapy was and continues to be a highly unpredictable art with regard to therapeutic effects. At the outset, I note that the Examiner provided no support for such a conclusion in connection with gene therapy treatment in general, and no support for such a conclusion in connection with gene therapeutic treatment of Alzheimer's disease, in particular. By 1996 gene therapy had progressed to the point where, the Goodman & Gilman's, a standard reference on therapeutics included a chapter entitled "Gene-Based Therapies." Eck et al., 1996 in Goodman and Gilman's: The Pharmacological Basis of Therapeutics (Ninth Edition), pages 77-101 (attached at Tab P). This chapter included a list of 58 clinical trials that had been approved by the NIH RAC between July 1990 and August 1994. *Id.* at 79-81. Thus, by 1996 gene therapy was sufficiently predictable to warrant human trials for a large number of diseases. Moreover, my own experience as a researcher and practitioner in the field of gene therapy for neurodegenerative diseases leads me to the opposite conclusion than the one reached by the Examiner. The introduction and widespread use of recombinant AAV vectors in about 1993 had particular relevance for neurological diseases. Through the application of standardized methods of vector packaging, purification and intracerebral delivery, by 1997 one of

ordinary skill in the art of gene therapy was able to successfully and reproducibly obtain expression of gene products in the brains of many species. *See, e.g.,* Kaplitt et al., *supra.*, Tab C and McCown et al., *supra.*, Tab M.

22. Thus, all of the technology for successful gene therapy in the brain was available in 1997. Moreover, as set out above, in my opinion, the '850 application provides a coherent and rational basis for using free end-specific antibodies to Aβ for the treatment of Alzheimer's disease and also provides sufficient information to enable a researcher working in the field of gene therapy in 1997 to practice the methods defined by the currently pending claims of this application. The '850 application further sets out detailed protocols for obtaining the free end-specific antibodies and their use in treating Alzheimer's disease. In short, by April 1997 it was entirely predictable by those skilled in the filed of gene therapy that free end-specific antibodies to Aβ could be used in gene therapy approaches to treat Alzheimer's disease. I am aware of no facts that would lead me to reach a contrary conclusion.

23. The Examiner's conclusion concerning the continuing unpredictability of gene therapy for neurological disorders is further refuted by the results of clinical trials that demonstrate gene therapy can be used to treat central nervous system disorders. Thus, gene therapy has been used in clinical trials to treat neurodegeneration. They include *in vivo* NGF gene therapy for Alzheimer's disease by Ceregene, *in vivo* Neurturin gene therapy (Sponsor: Ceregene) for Parkinson's Disease, *in vivo* Aromatic Amino Acid Decarboxylase (AADC; Sponsor: Genzyme), *in vivo* Glutamic Acide Decarboxylase ($GAD_{65/67}$; Sponsor: Neurologix) for Parkinson's Disease. Phase I trials using Neurturin, AADC and $GAD_{65/67}$ were all conducted using rAAV2 and have been completed without severe adverse effects. These have been presented to the RAC and at national meetings and provide evidence of efficacy. The results of these clinical trials demonstrate that Examiner's assertion that gene therapy continues to be unpredictable is not warranted.

24. In summary, for the reasons set forth above, it is my opinion that the '850 application provided sufficient information to enable one of ordinary skill in the art to use gene therapy in April 1997 to inhibit accumulation or neurotoxicity of Aβ by contacting soluble Aβ in the CSF of a patient suffering from Alzheimer's Disease with a free-end specific antibody to Aβ, as

called for in the claims pending in the subject patent application. No further information would have been required beyond what is disclosed in the '850 application to practice the claimed invention, nor would it have required undue experimentation.

25. I further declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements were made with the knowledge that willful false statements and the like so made are punishable by fine or imprisonment or both under Section 1001 of Title 18 of the United States Code, and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Declarant's signature:

Howard J. Federoff, M.D., Ph.D.

8/20/07

Date